



### **Review of retreat action items and status of sequences**

**1) Compile an on-line protocol bible for the three microorganisms.** This is a first priority because the next priority is to set up the local culture center.

**2) Compile all the sequences we need.** So far, we don't have a single full sequence except for G20 from JGI. Judy said she'd follow up with TIGR to get us the sequence. She said she'd send the old file that she has to the computational core. Joe similarly offered his oneidensis file. JGI already promised us the Geobacter file. I will follow up the TIGR sequences with Jonathan Eisen as another route. We will call in a week to update status.

**3) All the initial data types to be generated will be sent to Adam within a week.** This includes, a) the type of data to be generated, b) the protocol(s) used, c) the information about the experiment they want stored, d) the format of the files to be sent over. These formats are not binding but will be used in the initial designs of the data upload interfaces. Adam and Martin Keller will hash out the particular problems of the diverse proteomic data. (Adam will phone Martin later this week)

### **Discussion of web pages**

The investigators will fill out the investigator information page sent to them with these minutes. (Inna Dubchak's is sent as an example) All your permanent personnel should fill out these pages.

### **Discussion of OMICS paper**

We are going to submit a short overview to OMICS by Monday, October 28. Evidently, at least two other GTL projects have already submitted articles. Joe wanted to make sure we weren't too detailed or too broad with our ambitions. Adam said he would keep it high level and compliant with our overview. He will send out a draft at the end of the week for final comment. If the draft isn't deemed good enough we will not submit the article.

### **Discussion of the February GTL PI's meeting**

Adam reminded the team about the GTL PI's meeting in Washington, D.C. February 10-12, 2003. Judy voiced concern that Anna Palmisano is running a meeting from Feb 2-16<sup>th</sup> that overlaps with the GTL PI's meeting. She is helping with that meeting so may not be able to attend the GTL meeting. Adam will notify David Thomassen.

### **Discussion of the advisory panels**

The current technology advisory panel was reviewed. The panel includes Nielson, Frederickson, Lovely, and Tiedje. We need to recruit some bioinformaticists. Adam would like George Church and/or Jonathan Eisen and perhaps Andrey Rzhetsky. But other nominations are accepted.

We also need to assemble our scientific advisory panel and set a date for our next review.

## II. Budget Issues

### Review of contracts

All contracts seem to be in place with the exception of David's. The problem seems to be with his budget office. **We will track this down.**

### Discussion of the '03 budget and the congressional continuing resolution

DOE headquarters guidance mandates conservative spending until the federal budget is approved. Adam cautioned against front-loading costs and asked lab heads to project costs based on a 20% reduction in overall funds (10% per DOE guidance and 10% to be safe). Sub-contractors (U Wash, U Missouri, Diversa) will have their budgets allocated on a quarterly basis until we have our final budget. While we should act conservatively, we also need to get our operations going. Recruitments should be pursued, but term hires should be made wherever possible. Large purchases or commitments should be discussed with Adam first.

## III. Work Plans

Though the final formal workplans are not complete, each PI recounted their understanding of their work over the next six months. **Formal workplans will be delivered to Adam by early next week (September 30).** These will include a Gantt chart for each task in the project, a description of each task, and contact information for each task. An example of (perhaps an overly ambitious) Gantt chart is shown above and will be sent as an email attachment.

### Computational Core

Adam outlined the CC work plan from now up to the February PI's meeting. The goal is to have the three sequences in house, have an initial annotation accomplished and have a rough comparative genomics pipeline running with first stage operon and cis-regulatory predictions done. In addition, the data upload pages and flat-file database should be close to completed. The protocol repository will be part of this. Judy and David agreed to look into other sequences of related organisms we could look at to get a better comparative analysis, including G20 and the other Desulfito-bacteria. This is all pending receiving the sequences as described above. Initial prediction of the stress response pathways members will be made.

### Applied Environmental Microbiology Core

As Terry was just returning from travel and not able to be present, David outlined the AEMC plan roughly. The initial goals include: surveying the site [chemistry to ascertain the most important stressors and their ranges, doing an initial estimate of community structure and identifying the organisms at these sites most related to our targets. He will collect these and attempt to enrich. These may also be passed to Diversa or JGI for sequencing. The development of local culture facilities was brought up as a priority

(ultimately we may generate a central culture facility around the environmental simulation core). Judy has already sent Martin, Anup (and Massood), Jay and Joe samples of *Desulfovibrio*. Getting reproducibility of cultures among sites is a priority. This also bled over into the FGC work plan because it was not clear, especially in the absence of local culture facilities, how to prepare samples for molecular profiling without perturbing our responses. For example, chilling the sample or freezing it could cause stress responses we didn't control for. Judy, Anup, Jay and Joe will experiment with different methods.

Judy and David then had a brief discussion about whether or not *Desulfovibrio* could use sulfate as a terminal electron acceptor. They think not, since it accumulates during culture. They are also not sure if it can reduce Uranium. Judy may try this. But these have an impact on measuring oxidative stress response because the oxygen reacts with the disulfides produced making it difficult to control O<sub>2</sub> concentration (reference to Herbert Bianca, whose protocols Judy wants to look at, or perhaps she will visit his lab). David vetted the idea of growing the cells on pyruvate or syntrophically with a methanogen. David thought the latter interesting because it was a very simple community we could study and he thought the FGC could overcome the difficulties with mixed culture. He said it may be possible to do the proteomics of the community with Dick Smith, for example.

At this point the choice of the first stressors was briefly discussed: O<sub>2</sub>, osmotic stress, heat shock, and pH were all considered. Since Joe is already working on O<sub>2</sub>, pH and heat shock in *Shewanella*, these seem to be the natural for *Desulfovibrio*. Heat shock is the best understood pathway, so it will form the baseline for the analysis.

### **Functional Genomics Core**

Jay, Judy and Anup outlined the FGC work plan. The mass spec machines have arrived and Jay expects to develop and test protocols for metabolomics, first on *E. coli* and *B. subtilis* and then on *Desulfovibrio* once we have culture conditions working. Anup and Massood are working on the culturing for their proteomics aspect and Adam will get the low-down from Martin about his proteomics effort. Joe will transport *Shewanella* data to Adam in the next couple of weeks. Jay will meet with Anup to see about interface to Anup's CE technology with the mass specs at Berkeley. The FGC will try to get all three data types: protein expression, gene expression, and metabolite expression in its very initial form, in some organism in the first semester of the grant.

